REMARKS

Claims 66-70, 74, 78, 80, 84-88, 93, 97-101, and 105-145 are pending and stand rejected. Applicants have herein amended claims 133-145 and added claims 146-150. Support for the amendments and new claims may be found throughout the specification, e.g., at pages 5, 7-8, 15, 20, and 27, and in prior versions of the claims. Accordingly, no new matter has been added. Thus, claims 66-70, 74, 78, 80, 84-88, 93, 97-101, and 105-150 are pending.

In light of the amendments and the remarks herein, Applicants respectfully request reconsideration and allowance of claims 66-70, 74, 78, 80, 84-88, 93, 97-101, and 105-150.

Objections to the Specification

The Examiner objected to the specification since the priority data in the first line of the specification did not claim priority to U.S. Application No. 08/878,801. Applicants have herein amended the specification to recite the suggested priority information. Accordingly, Applicants respectfully request withdrawal of the objection.

Claim Objections

The Examiner objected to the syntax of claims 139-145 and suggested a re-organization and combining of certain claim limitations. Applicants have herein amended claims 139-145 per some of the Examiner's suggestions - namely, combining the limitations of step (iii) (regarding COS-7 cells and a CMV inducible promoter) with step (1).

The Examiner also suggested that the limitation in step (c) reciting "wherein induced expression of said Gα15 is sufficient to permit promiscuous coupling to said GPCR" should be removed as inherent in the prior limitation (stating that a 3-fold increase in Gα15 expression is required). Applicants respectfully disagree, as a 3-fold increase in Gα15 expression may not necessarily produce this coupling. Accordingly, Applicants respectfully suggest that this deletion would be inappropriate, and therefore they have not deleted the limitation.

Given the above, Applicants respectfully suggest that the syntax of the present claims has been improved by the amendments herein, and respectfully request withdrawal of the claim objections.

Rejections under 35 U.S.C. § 112, second paragraph

The Examiner rejected claims 66-70, 74, 78, 80, 84-88, 93, 97-101, and 105-145 under 35 U.S.C. § 112, second paragraph, as being indefinite for failing to particularly point out and distinctly claim the subject matter which applicant regards as the invention. The Examiner then set forth particular comments in paragraphs (B)-(J), to which Applicants hereby respond in turn.

In paragraph (B), the Examiner rejected claims 66-70, 81, 84-88, 97-101, 105-109, 139, 142, 144, and 145 as confusing, since it was not clear to the Examiner how a reporter gene can be used to detect activation of the GPCR if a reporter gene substrate was not required until claims 110, 112, 114, and 116. Applicants respectfully point out that not all polypeptide products of reporter genes require a substrate for detection. For example, GFP is a naturally fluorescent protein that permits detection using fluorescence-based techniques. See, for example, page 15 of the specification, wherein a receptor gene is defined as a polynucleotide encoding a protein readily detectable by its presence or activity. Accordingly, Applicants respectfully request withdrawal of the rejections.

In paragraph (C), the Examiner rejected claims 68-70, 86-88, 99-101, and 107-109 as confusing, since it was not clear to the Examiner the purpose of increasing calcium levels in the cells or for using PMA, and that nothing in the independent claims appeared to require, or depend on, increased calcium levels or PMA. Applicants refer to page 20 of the specification, where it states that "activation of Gα15 . . . can, through a G-protein signaling pathway, activate PLCβ, which in turn increases intracellular calcium levels. An increase in calcium levels can lead to a modulation of a calcium-responsive promoter," e.g., a calcium-responsive promoter such as NFAT that is operably linked to a reporter gene. On pages 23-25, the specification notes that it may be preferable to increase intracellular calcium levels to a "subthreshold level" of activation of the regulatory control sequence linked to the reporter gene. The reporter gene is thus poised for activation by increase of the intracellular concentration of calcium to a subthreshold level of activation, so that if coupling of a Gα15 protein to a GPCR occurs, PLCβ is activated, which in turn subsequently *further* increases intracellular calcium levels to *above-threshold* levels, with concomitant enhanced expression of a reporter gene. Indeed, the specification states on page 24:

> For example, in order to enhance detection of expression of a reporter gene, the cell can be contacted with a compound (e.g., a calcium ionophore) that increases calcium levels inside the cell. By increasing calcium levels inside the cell, the probability that activation of a G-protein will activate the expression of a reporter gene is greatly enhanced. Preferably, the calcium levels are increased to a level that is just below the threshold level for activation of a calcium responsive promoter such as an NFAT promoter.

Similarly, the specification notes that protein kinase C can be activated to this "subthreshold" level with PMA; see, e.g., page 25.

Given the above, Applicants respectfully assert that claims 68-70, 86-88, 99-101, and 107-109 are therefore not confusing, since the purpose of increasing calcium levels or for using PMA is clearly explained in the specification. Accordingly, Applicants respectfully request withdrawal of the rejections.

In paragraph (D), the Examiner rejected claims 74, 78, 93, 126, 127, 128 as confusing, since the purpose of an intracellular calcium indicator or how the indicator related to the claimed methods was not understood. Given the discussion above with respect to paragraph (C), and also given the statements in the specification on page 27, Applicants submit that the intracellular calcium indicator's purpose is not confusing. The intracellular calcium indicator is used to detect a change in calcium levels in a cell, e.g., as a result of activation of PLCβ by coupling of a Gα protein to a GPCR. Thus, an intracellular calcium indicator can be used a detection method in the present claims. Applicants also refer the Examiner to the Molecular Probes (Eugene, OR) web page (www.molecularprobes.com) and/or catalog; Molecular Probes sells various calcium indicators, including Fura II (e.g., as recited in claims 126-128). Accordingly, Applicants respectfully request withdrawal of the rejections.

In paragraph (E), the Examiner rejected claims 80, 84, 97, and 105 as confusing, asserting it was not clear what process in the independent claims would lead to a change in fluorescence. Applicants refer to pages 15-19 of the specification, wherein it is noted that a reporter gene can produce, for example, a fluorescent polypeptide such as GFP or produce a fluorescent product from a substrate. In addition, some of the independent claims recite that a signal transduction detection system can comprise a dye. The specification describes dyes on page 8 as molecules that absorb light, including ultraviolet light, and sets forth fluorescent dyes on page 9. In addition, numerous other dyes that fluoresce are known to those of skill in the art

and can be used in the detection methods of the present invention. Accordingly, Applicants respectfully request withdrawal of the rejections.

In paragraph (F), the Examiner rejected claims 139-145 as confusing, since the recitation "first heterologous inducible promoter" gave the impression of more than one inducible promoter. Claims 139-145 have been amended herein consistently with the Examiner's suggestions, thereby obviating the rejections.

In paragraph (G), the Examiner rejected claims 139-145 as confusing with respect to a tet-dependent transactivator linked to a constitutive promoter, which did not appear to be required for the invention. Applicants refer to page 19 of the specification; to pages 4-6 of the Office Action dated 8/22/02; and to pages 3-4 of the Office Action dated 10/24/2002. In particular, the "Conclusion" on page 4 of the Office Action dated 10/24/2002 indicates that claims 139-145, which recite that the cells comprise a polynucleotide encoding a tet-dependent transactivator linked to a constitutive promoter, would be allowable if rewritten in independent format to include all limitations of the claims from which they depend. Without acquiescing in the prior enablement rejections under 35 U.S.C. § 112, first paragraph, and without prejudice to further prosecution of claims that do not recite such a limitation, Applicants have herein maintained the previously-introduced limitation in order to expedite prosecution of the pending claims. Accordingly, Applicants respectfully request withdrawal of the rejections.

In paragraph (H), the Examiner rejected claims 139-145 as confusing with respect to the requirement of the claimed cell line regarding "arising from functional cell analysis." In order to clarify the claims, Applicants have herein amended the claims to delete the limitation regarding "arising from functional cell analysis." Accordingly, Applicants respectfully request withdrawal of the rejections.

In paragraph (I), the Examiner rejected claim 145 as being incomplete for omitting essential steps, namely omitting a control step as recited in dependent claims 133-138. Applicants submit that amended claim 145, which relates to functional profiling, does not need a control step as recited in claims 133-138, because step (iii) involves a comparison among different clones in the panel. Thus, there is an internal comparison of relative levels of expression, rather than a control. In addition, Applicants note that in new claim 146 such a control step is recited. Accordingly, Applicants respectfully request withdrawal of the rejection.

In paragraph (J), the Examiner rejected claims 139-145 as confusing, since it was not clear for which protein the first heterologous inducible promoter provided low level expression. As suggested by the Examiner, claims 139-145 have been amended herein to recite low level expression of Gα15. Accordingly, Applicants respectfully request withdrawal of the rejections.

CONCLUSION

In light of the amendments and the remarks herein, Applicants respectfully assert that all claims are in condition for allowance, which action is requested. The Examiner is invited to call the under-signed attorney if such would expedite prosecution.

Enclosed is a $ 420 check for the Petition for Extension of Time fee. Please apply any other charges or credits to deposit account 06-1050.

Respectfully submitted,

Date: 10/2/03

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